

Docket No. 50046290-0007

Serial No. To Be Assigned

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This Application claims priority to PCT Application No. PCT/US00/14818, filed May 26, 2000, which claims priority to United States Application No. 09/322,700, filed May 28, 1999, now U.S. Patent No. 6,172,040.--

In the Claims

Please cancel claims 50 and 52-55.

Please amend the claims as follows:

A²

4. (Amended) The composition in accordance with claim 1, wherein the naturally occurring substrate not including gelatin is a galactose-rich polysaccharide comprising mainly galactose residues and derivatized galactose residues.

A³

6. (Amended) A composition of matter comprising a dispersion of isolated lactoferrin immobilized on a naturally occurring substrate via the N-terminus region of the lactoferrin, and native lactoferrin.

A⁴

10. (Amended) The composition in accordance with claim 6, wherein the composition comprises about 1 % wt/vol immobilized lactoferrin and about 1 % wt/vol native lactoferrin.

A⁵

14. (Amended) A composition of matter comprising an aqueous buffer solution containing a physiologically acceptable acid selected from the group consisting of oxalic acid, ethylenediamine tetraacetic acid, carbonic acid, and citric acid; a physiologically

AS CONCLUDED - acceptable base; and a physiologically acceptable salt selected from the group consisting of calcium chloride, potassium chloride, and sodium chloride, wherein the ratio of acid to base to salt is 0.1 to 0.0001M (acid) : 1 to 0.001M (base) : 10 to 0.01M (salt) and containing a mixture of native lactoferrin and isolated lactoferrin immobilized on a galactose-rich polysaccharide comprising mainly galactose residues and derivatized galactose residues, collagen, gelatin, fibronectin, casein, mucin, heparan-sulfate, carrageenan, deoxyribonucleic acid, adenosine triphosphate or a triglyceride via the N-terminus region of the lactoferrin, in a native lactoferrin to isolated immobilized lactoferrin molar ratio of from about 1:1 to about 1:5 and in a concentration of from about 0.001 to about 2.5 % wt/vol.

18. (Amended) A method for reducing the microbial contamination of a composition subject to microbial contamination by a microbe, comprising: treating the composition with a sufficient amount of isolated lactoferrin immobilized on a naturally occurring substrate via the N-terminus region of the lactoferrin to reduce microbial contamination.

40. (Amended) A method for inhibiting the microbial contamination of a composition subject to microbial contamination comprising treating the composition with an aqueous buffer solution containing a physiologically acceptable acid selected from the group consisting of oxalic acid, ethylenediamine tetraacetic acid, carbonic acid, and citric acid; a physiologically acceptable base; and a physiologically acceptable salt selected from the group consisting of calcium chloride, potassium chloride, and sodium chloride, wherein the ratio of acid to base to salt is 0.1 to 0.0001M (acid): 1 to 0.001M (base): 10 to 0.01M (salt) and containing a mixture of native lactoferrin and isolated lactoferrin immobilized on a

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galactose-rich polysaccharide comprising mainly galactose residues and derivatized galactose residues, collagen, gelatin, fibronectin, casein, mucin, heparan-sulfate, carrageenan, deoxyribonucleic acid, adenosine triphosphate or a triglyceride via the N-terminus region of the lactoferrin, in a native lactoferrin to isolated immobilized lactoferrin molar ratio of from about 1:1 to about 1:5 and in a concentration of from about 0.001 to about 2.5 % wt/vol.

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51. (Amended) The method in accordance with claim 40, wherein the ratio of acid to base to salt is 0.01 to 0.001M (acid) : 0.1 to 0.01M (base) : 1 to 0.1M (salt).

A⁹
70. (Amended) A method for reducing the microbial contamination of a meat product subject to microbial contamination by a microbe, comprising: applying to the meat product a composition containing a physiologically acceptable acid selected from the group consisting of oxalic acid, ethylenediamine tetraacetic acid, carbonic acid, and citric acid; a physiologically acceptable base; and a physiologically acceptable salt selected from the group consisting of calcium chloride, potassium chloride, and sodium chloride, wherein the molar ratio of acid to base to salt is 0.1 to 0.0001 (acid) : 1 to 0.001 (base) : 10 to 0.01 (salt) and containing a mixture of native lactoferrin and isolated lactoferrin immobilized on a galactose-rich polysaccharide comprising mainly galactose residues and derivatized galactose residues, collagen, gelatin, fibronectin, casein, mucin, heparan-sulfate, carrageenan, deoxyribonucleic acid, adenosine triphosphate or a triglyceride via the N-terminus region of the lactoferrin, in a native lactoferrin to isolated immobilized lactoferrin molar ratio of from about 1:1 to about 1:5 and in a concentration of from about 0.001 to about 2.5 % wt/vol.

~~1426~~ ~~80~~
~~83.~~ (Amended) The method of claim 82, wherein species is *Clostridium*
perfringens, *Clostridium difficile*, *Clostridium botulinum*, or *Clostridium tetani*.

Please add the following claims:

~~1426~~ ~~102~~
~~105.~~ A composition of matter comprising a dispersion of isolated lactoferrin
 immobilized on a naturally occurring substrate via the N-terminus region of the lactoferrin;
 and at least one pharmaceutically acceptable carrier.

~~103~~
~~106.~~ The composition in accordance with claim 105, wherein the naturally
 occurring substrate is a protein, a polysaccharide, cellulose, a nucleic acid, a nucleotide, or a
 lipid.

~~104~~
~~107.~~ The composition in accordance with claim 105, wherein the naturally
 occurring substrate is collagen, gelatin, fibronectin, casein, mucin, heparan-sulfate,
 carrageenan, deoxyribonucleic acid, adenosine triphosphate or a triglyceride.

~~105~~
~~108.~~ The composition in accordance with claim 105, wherein the naturally
 occurring substrate is a galactose-rich polysaccharide comprising mainly galactose residues
 and derivatized galactose residues.

~~106~~
~~109.~~ The composition of claim 105, wherein the dispersion is an aqueous solution,
 an aqueous emulsion, a colloid, a suspension, a powder, or a granular solid.

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110. The composition in accordance with claim 105, further comprising native lactoferrin.

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111. The composition in accordance with claim 110, wherein the concentration of immobilized lactoferrin and native lactoferrin in the dispersion is from about 0.05% wt/vol to about 2.5 % wt/vol.

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112. The composition in accordance with claim 110, wherein the molar ratio of immobilized lactoferrin to native lactoferrin is a ratio of from about 1:1 to about 1:10.

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113. The composition in accordance with claim 110, wherein the molar ratio of immobilized lactoferrin to native lactoferrin is a ratio of from about 1:1 to about 1:5.

111
114. The composition in accordance with claim 110, wherein the composition comprises about 1 % wt/vol immobilized lactoferrin and about 1 % wt/vol native lactoferrin.

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115. The composition in accordance with claim 110, wherein the composition further comprises a buffer system.

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116. The composition in accordance with claim 115, wherein the buffer system contains a physiologically acceptable acid, a physiologically acceptable base, and a physiologically acceptable salt.

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117. The composition in accordance with claim 116, wherein the physiologically acceptable acid is oxalic acid, ethylenediamine tetraacetic acid, carbonic acid, or citric acid;

the physiologically acceptable base is sodium bicarbonate, potassium bicarbonate, sodium carbonate, or potassium carbonate; and the physiologically acceptable salt is calcium chloride, potassium chloride or sodium chloride.

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118. The composition of claim 105, wherein the carrier is selected from the group consisting of solid, semisolid or liquid glucose, lactose, sucrose, gum acacia, agar, petrolatum, lanolin, dimethyl sulfoxide, normal saline, phosphate buffered saline, sodium alginate, bentonite, carbomer, carboxymethylcellulose, carageenan, powdered cellulose, cholesterol, gelatin, hydroxyethyl cellulose, hydroxypropyl cellulose, hydroxypropyl methylcellulose, methylcellulose, octoxynol 9, oleyl alcohol, polyvinyl alcohol, povidone, propylene glycol monostearate, sodium lauryl sulfate, sorbitan esters, stearyl alcohol, tragacanth, xanthan gum, chondrus, glycerin, trolamine, avocado oil, almond oil, coconut oil, coconut butter, propylene glycol, ethyl alcohol, malt, and malt extract.

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119. The composition of claim 105, further comprising a pharmaceutically acceptable emulsifier.

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120. The composition of claim 119, wherein the emulsifier is selected from the group consisting of monoglyceride compounds, diglyceride compounds, glycerol, phosphatidyl ethanolamine, phosphatidyl choline, or lecithin.

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121. The composition in accordance with claim 14, wherein the molar ratio of acid to base to salt is 0.01 to 0.001M (acid) : 0.1 to 0.01M (base) : 1 to 0.1M(salt).

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~~122~~. The composition of claim 105, wherein the composition is formulated in a cosmetic, a cleanser, a food supplement, or a medicament.

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~~123~~. The composition of claim 122, wherein the cosmetic, cleanser, food supplement, or medicament is formulated for applying to an external surface of a vertebrate subject.

111 CONT.
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~~124~~. The composition of claim 123, wherein the vertebrate subject is a human.

¹²²
~~125~~. The composition of claim 123, wherein the vertebrate subject is a non-human vertebrate.

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~~126~~. The composition of claim 122, wherein the cleanser is formulated as a pharmaceutically acceptable skin cleanser.

¹²⁴
~~127~~. The composition of claim 122, wherein the medicament is formulated in a pharmaceutically acceptable delivery system.

¹²⁵
~~128~~. The composition of claim 127, wherein said delivery system is an injection, intravenous drip, inhalant, or implant delivery system.

¹²⁶
~~129~~. The composition of claim 127, wherein said delivery system is a transdermal delivery system.

Docket No. 50046290-0007

Serial No. To Be Assigned

9

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~~130~~. The composition of claim 127, wherein said delivery system is a transmucosal delivery system.

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~~131~~. The composition of claim 127, wherein said delivery system is an oral transmucosal delivery system.

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~~132~~. The composition of claim 127, wherein said delivery system is a vaginal transmucosal delivery system.

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~~133~~. The composition of claim 127, wherein said delivery system comprises an adhesive patch.

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~~134~~. The composition of claim 127, wherein said delivery system comprises a gel, cream, ointment, suppository, sanitary wipe, bandage, or shampoo.

132
~~135~~. The composition of claim 127, wherein the delivery system is a mouth wash, gargle solution, denture cleanser, or dentifrice.

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~~136~~. The composition of claim 127, wherein the delivery system is a toothpaste or chewing gum.

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~~137~~. The composition of claim 127, wherein the medicament is formulated in a urogenital, rectal, or colonic delivery system.

Docket No. 50046290-0007

Serial No. To Be Assigned

10

R.126
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138. The composition of claim 122, wherein the composition further comprises an antibiotic or probiotic agent.

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139. The composition of claim 127, wherein the delivery system is a suppository, gel, or foam.

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140. The composition of claim 127, wherein the medicament is formulated in an ingestive delivery system.

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141. The composition of claim 140, wherein the ingestive delivery system is a tablet, capsule, caplet, troche, lozenge, coated or uncoated microspheres or particles, dispersible powder or granules, syrup, elixir, beverage, or food additive.

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142. The composition of claim 141, wherein the tablet or capsule comprises a controlled release coating.

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143. The composition of claim 141, wherein the ingestive delivery system comprises an enteric coating to prevent esophageal or gastric release of immobilized lactoferrin.

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144. The composition of claim 127, wherein the delivery system comprises a lavage or enema.

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~~145.~~ The composition of claim 122, wherein the medicament is formulated for treating a human.

~~143~~
~~146.~~ The composition of claim 145, wherein the composition is formulated for pediatric use.

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~~147.~~ The composition of claim 122, wherein the medicament is formulated for veterinary use.

~~145~~
~~148.~~ The composition of claim 147, wherein the composition is formulated for use in a domestic or farm animal.

~~146~~
~~149.~~ The composition of claim 147, wherein the composition is formulated for use in a non-human mammal or bird.

~~147~~
~~150.~~ The composition of claim 149, wherein the composition is formulated for use in a non-human primate, mouse, rat, rabbit, gerbil, hamster, canine, feline, ovine, bovine, swine, pachyderm, equine, or marine mammal.

~~148~~
~~151.~~ The composition of claim 149, wherein the composition is formulated for use in a chicken, duck, goose, turkey, ostrich, emu, dove, pigeon, quail, pheasant, peafowl, or guinea fowl.

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~~152.~~ The method of claim 18, wherein said composition subject to microbial contamination is a human.

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~~153.~~ The method of claim 152, wherein treating includes administering to said human said composition by a pharmaceutically acceptable delivery route.

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~~154.~~ The method of claim 153, wherein said delivery route is non-systemic.

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~~155.~~ The method of claim 154, wherein said non-systemic delivery route is a urogenital, rectal, or colonic delivery route.

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~~156.~~ The method of claim 154, wherein said non-systemic delivery route is a topical application of a cream, gel, or ointment.

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~~157.~~ The method of claim 153, wherein said delivery route is systemic.

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~~158.~~ The method of claim 157; wherein said systemic delivery route is by ingestion, injection, intravenous drip, inhalant, or implant.

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~~159.~~ The method of claim 157 wherein said systemic delivery route is a transdermal delivery route.

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~~160.~~ The method of claim 157 wherein said systemic delivery route is a transmucosal delivery route.

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~~161.~~ The method of claim 152, wherein the microbial contamination of a human to be reduced is in the gastrointestinal system of the human.

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~~162.~~ The method of claim 153, wherein treating further comprises administering an antimicrobial agent or probiotic agent in conjunction with the immobilized lactoferrin.

11 cont.
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~~163.~~ The method of claim 162, wherein the probiotic agent is a species of *Bifidobacterium*, *Streptococcus*, *Pediococcus*, *Lactococcus*, or *Lactobacillus*.

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~~164.~~ The method of claim 163, wherein the probiotic agent is *Bifidobacterium bifidum*, *Bifidobacterium longum*, *Bifidobacterium animalis*, *Streptococcus lactis*, *Streptococcus cremoris*, *Streptococcus thermophilus*, *Pediococcus pentoseus*, *Lactococcus lactis*, *Lactobacillus acidophilus*, *Lactobacillus rhamnosus*, *Lactobacillus plantarum*, *Lactobacillus reuteri*, *Lactobacillus bulgaricus*, *Lactobacillus paracasei*, or *Lactobacillus casei*.

¹⁶²
~~165.~~ The method of claim 162, wherein the antimicrobial agent is an antibiotic.

¹⁶³
~~166.~~ The method of claim 165, wherein the antimicrobial agent is neomycin, metronidazole, teicoplanin, vancomycin, ciprofloxacin, doxycycline, tetracycline, augmentin, erythromycin, chloramphenicol, cephalexin, penicillin, ampicillin, kanamycin, rifamycin, rifaximin, rifampin, clindamycin, trimethoprim, a4-amino salicylate compound, a 5-aminosalicylate compound, a sulfonamide compound, a betalactam compound, an aminoglycoside compound, a macrolide compound, or a quinolone compound.

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167. The method of claim 152, wherein the microbe is bacterium, a fungus, a protozoan, or a virus.

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168. The method in accordance with claim 152, wherein the microbe is enterotoxigenic *Escherichia coli*, enteropathogenic *Escherichia coli*, *Shigella dysenteriae*, *Shigella flexneri*, *Salmonella typhimurium*, *Salmonella typhi*, *Salmonella abony*, *Salmonella dublin*, *Salmonella enteritidis*, *Salmonella hartford*, *Salmonella kentucky*, *Salmonella panama*, *Salmonella pullorum*, *Salmonella rostock*, *Salmonella thompson*, *Salmonella virchow*, *Enterobacter aerogenes*, *Vibrio cholerae*, *Yersinia enterocolitica*, *Campylobacter jejuni*, *Aeromonas hydrophila*, *Staphylococcus aureus*, *Staphylococcus hyicus*, *Staphylococcus epidermidis*, *Staphylococcus hominis*, *Staphylococcus warneri*, *Staphylococcus xylosus*, *Staphylococcus chromogenes*, *Streptococcus pyogenes*, *Streptococcus pneumoniae*, *Streptococcus mutans*, *Streptococcus sanguis*; *Pediococcus acne*, *Bacillus cereus*, *Bacillus anthracis*, *Bacillus subtilis*, a *Brucella species*, *Listeria monocytogenes*, *Legionella pneumophila*, *Bordetella pertussis*, *Pseudomonas aeruginosa*, *Legionella pneumophila*, *Francisella tularensis*, *Candida albicans*, *Brochothrix thermospacta*, *Bacillus pumilus*, *Enterococcus faecium*, *Actinobacillus actinomycetemcomitans*, *Porphyromonas gingivalis*, *Prevotella intermedia*, *Deinococcus radiopugnans*, *Deinococcus radiodurans*, *Deinobacter grandis*, *Acinetobacter radioresistens*, or *Methylobacterium radiotolerans*.

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169. The method in accordance with claim 152, wherein the microbe is a verotoxic *Escherichia coli*.

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~~170.~~ The method in accordance with claim 169, wherein the verotoxic *Escherichia coli* is the serotype O157:H7.

165
171. The method of claim 152, wherein the microbe is a *Clostridium* species.

~~472.~~ ¹⁶⁷ The method of claim 171, wherein species is *Clostridium perfringens*, *Clostridium difficile*, *Clostridium botulinum*, or *Clostridium tetani*.

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173. The method of claim 152, wherein the microbe is a protozoan selected from
the group consisting of *Entamoeba histolytica*, *Naegleria fowleri*, *Giardia lamblia*,
Leishmania spp., *Trichomonas vaginalis*, *Trypanosoma spp.*, *Plasmodium spp.*, or
Taxoplasrna spp.

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~~174.~~ The method of claim 18, wherein said composition subject to microbial contamination is a non-human vertebrate.

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~~175.~~ The method of claim 174, wherein treating includes administering to said non-human vertebrate said composition by a pharmaceutically acceptable delivery route.

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176. The method of claim 175, wherein said delivery route is non-systemic.

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~~177~~. The method of claim 176, wherein said non-systemic delivery route is a urogenital, rectal, or colonic delivery route.

R126
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~~178.~~ The method of claim 176, wherein said non-systemic delivery route is a topical application of a cream, gel, or ointment.

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~~179.~~ The method of claim 175, wherein said delivery route is systemic.

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~~180.~~ The method of claim 179, wherein said systemic delivery route is by ingestion, injection, intravenous drip, inhalant, or implant.

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~~181.~~ The method of claim 179, wherein said systemic delivery route is a transdermal delivery route.

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~~182.~~ The method of claim 179, wherein said systemic delivery route is a transmucosal delivery route.

~~180~~
~~183.~~ The method of claim 174, wherein the microbial contamination of a non-human vertebrate to be reduced is in the gastrointestinal system of the non-human vertebrate.

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~~184.~~ The method of claim 175, wherein treating further comprises administering an antimicrobial agent or probiotic agent in conjunction with the immobilized lactoferrin.

~~182~~
~~185.~~ The method of claim 184, wherein the probiotic agent is a species of *Bifidobacterium*, *Streptococcus*, *Pediococcus*, *Lactococcus*, or *Lactobacillus*.

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186.

The method of claim 185, wherein the species is *Bifidobacterium bifidum*, *Bifidobacterium longum*, *Bifidobacterium animalis*, *Streptococcus lactis*, *Streptococcus cremoris*, *Streptococcus thermophilus*, *Pediococcus pentoseus*, *Lactococcus lactis*, *Lactobacillus acidophilus*, *Lactobacillus rhamnosus*, *Lactobacillus plantarum*, *Lactobacillus reuteri*, *Lactobacillus bulgaricus*, *Lactobacillus paracasei*, or *Lactobacillus casei*.

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187.

The method of claim 184, wherein the antimicrobial agent is an antibiotic.

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The method of claim 187, wherein the antimicrobial agent is neomycin, metronidazole, teicoplanin, vancomycin, ciprofloxacin, doxycycline, tetracycline, augmentin, erythromycin, chloramphenicol, cephalexin, penicillin, ampicillin, kanamycin, rifamycin, rifaximin, rifampin, clindamycin, trimethoprim, a 4-amino salicylate compound, a 5-aminosalicylate compound, a sulfonamide compound, a betalactam compound, an aminoglycoside compound, a macrolide compound, or a quinolone compound.

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The method of claim 174, wherein the microbe is a bacterium, a fungus, a protozoan, or a virus.

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The method in accordance with claim 174, wherein the microbe is enterotoxigenic *Escherichia coli*, enteropathogenic *Escherichia coli*, *Shigella dysenteriae*, *Shigella flexneri*, *Salmonella typhimurium*, *Salmonella typhi*, *Salmonella abony*, *Salmonella dublin*, *Salmonella enteritidis*, *Salmonella hartford*, *Salmonella kentucky*, *Salmonella panama*, *Salmonella pullorum*, *Salmonella rostock*, *Salmonella thompson*, *Salmonella virchow*, *Enterobacter aerogenes*, *Vibrio cholerae*, *Yersinia enterocolitica*, *Campylobacter*

jejuni, Aeromonas hydrophila, Staphylococcus aureus, Staphylococcus hyicus, Staphylococcus epidermidis, Staphylococcus hominis, Staphylococcus warneri, Staphylococcus xylosus, Staphylococcus chromogenes, Streptococcus pyogenes, Streptococcus pneumoniae, Streptococcus mutans, Streptococcus sanguis; Pediococcus acne, Bacillus cereus, Bacillus anthracis, Bacillus subtilis, a Brucella species, Listeria monocytogenes, Legionella pneumophila, Bordetella pertussis, Pseudomonas aeruginosa, Legionella pneumophila, Francisella tularensis, Candida albicans, Brochothrix thermospacta, Bacillus pumilus, Enterococcus faecium, Actinobacillus actinomycetemcomitans, Porphyromonas gingivalis, Prevotella intermedia, Deinococcus radiopugnans, Deinococcus radiodurans, Deinobacter grandis, Ainetobacter radio resistens, or Methylobacterium radiotolerans.

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191.

The method in accordance with claim 174, wherein the microbe is a verotoxic *Escherichia coli*.

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The method in accordance with claim 191, wherein the verotoxic *Escherichia coli* is the serotype O157:H7.

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193.

The method of claim 174, wherein the microbe is a *Clostridium* species.

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The method of claim 193, wherein species is *Clostridium perfringens, Clostridium difficile, Clostridium botulinum, or Clostridium tetani*.

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195.

The method of claim 174, wherein the microbe is a protozoan selected from the group consisting of *Entamoeba histolytica*, *Naegleria fowleri*, *Giardia lamblia*, *Leishmania spp.*, *Trichomonas vaginalis*, *Trypanosoma spp.*, *Plasmodium spp.*, or *Taxoplasrna spp.*

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The method of claim 174, wherein said non-human vertebrate is a domestic or farm animal.

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The method of claim 174, wherein said non-human vertebrate is a mammal or bird.

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198.

The method of claim 174, wherein said non-human vertebrate is a non-human primate, mouse, rat, rabbit, gerbil, hamster, canine, feline, ovine, bovine, swine, pachyderm, equine, or marine mammal.

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The method of claim 174, wherein said non-human vertebrate is a chicken, duck, goose, turkey, ostrich, emu, dove, pigeon, quail, pheasant, peafowl, or guinea fowl.

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The method of claim 18, wherein said composition subject to microbial contamination is a biological surface or a biological fluid.

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201.

The method of claim 200, wherein the fluid is a culture medium.

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The method of claim 200, wherein the biological surface or fluid is in vitro.

Docket No. 50046290-0007

Serial No. To Be Assigned

20

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~~203.~~ The method of claim 200, wherein the biological surface is a cell surface, membrane surface, mucosal surface, epithelial surface, luminal surface, skin surface, or eggshell surface.

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~~204.~~ The method of claim 200, wherein the biological surface is an epithelial or mucosal surface.

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~~205.~~ The method of claim 200, wherein the biological fluid is semen, blood, lymph, urine, prostatic fluid, saliva, gastric juice, mucus, synovial fluid, pleural exudate, peritoneal exudate, pericaridal exudate, or cerebro-spinal fluid.

Applicant awaits a first office action on the merits.

Respectfully submitted,

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November 28, 2001

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